CLAIMS

1. A method for *in vivo* down-regulation of amyloid protein in an animal, including a human being, the method comprising effecting presentation to the animal's immune system of an immunogenically effective amount of

- at least one amyloidogenic polypeptide or subsequence thereof which has been formulated so that immunization of the animal with the amyloidogenic polypeptide or subsequence thereof induces production of antibodies against the amyloidogenic polypeptide, and/or

- at least one analogue of the amyloidogenic polypeptide wherein is introduced at least one modification in the amino acid sequence of the amyloidogenic polypeptide which has as a result that immunization of the animal with the analogue induces production of antibodies against the amyloidogenic polypeptide.

2. The method according to claim 1, wherein is presented an analogue with at least one modification of the amino acid sequence of the amyloidogenic polypeptide.

3. The method according to claim 2, wherein the modification has as a result that a substantial fraction of B-cell epitopes of the amyloidogenic polypeptide are preserved and that

- at least one foreign T helper lymphocyte epitope ($T_H$ epitope) is introduced, and/or

- at least one first moiety is introduced which effects targeting of the modified molecule to an antigen presenting cell (APC) or a B-lymphocyte, and/or

- at least one second moiety is introduced which stimulates the immune system, and/or

- at least one third moiety is introduced which optimizes presentation of the modified amyloidogenic polypeptide to the immune system.

4. The method according to claim 3, wherein the modification includes introduction as side groups, by covalent or non-covalent binding to suitable chemical groups in the amyloidogenic polypeptide or a subsequence thereof, of the foreign $T_H$ epitope and/or of the first and/or of the second and/or of the third moiety.

5. The method according to claim 3 ~~or 4~~, wherein the modification includes amino acid substitution and/or deletion and/or insertion and/or addition.

6. The method according to claim 5, wherein the modification results in the provision of a fusion polypeptide.

7. The method according to claim 5 ~~or 6~~, wherein introduction of the amino acid substitution and/or deletion and/or insertion and/or addition results in a substantial preservation of the overall tertiary structure of the amyloidogenic polypeptide.

8. The method according to ~~any one of claims 2-7~~ Claim 2, wherein the modification includes duplication of at least one B-cell epitope of the amyloidogenic polypeptide and/or introduction of a hapten.

9. The method according to ~~any one of claims 3-8~~ Claim 3, wherein the foreign T-cell epitope is immunodominant in the animal.

10. The method according to ~~any one of claims 3-9~~ Claim 3, wherein the foreign T-cell epitope is promiscuous, such as a foreign T-cell epitope which is selected from a natural promiscuous T-cell epitope and an artificial MHC-II binding peptide sequence.

11. The method according to claim 10, wherein the natural T-cell epitope is selected from a Tetanus toxoid epitope such as P2 or P30, a diphtheria toxoid epitope, an influenza virus hemagluttinin epitope, and a *P. falciparum* CS epitope.

12. The method according to ~~any one of claims 3-11~~ Claim 3, wherein the first moiety is a substantially specific binding partner for a B-lymphocyte specific surface antigen or for an APC specific surface antigen such as a hapten or a carbohydrate for which there is a receptor on the B-lymphocyte or the APC.

13. The method according to ~~any one of claims 3-12~~ Claim 3, wherein the second moiety is selected from a cytokine such as interferon γ (IFN-γ) or an effective part thereof, Flt3L or an effective part thereof, interleukin 1 (IL-1) or an effective part thereof, interleukin 2 (IL-2) or an effective part thereof, interleukin 4 (IL-4) or an effective part thereof, interleukin 6 (IL-6) or an effective part thereof, interleukin 12 (IL-12) or an effective part thereof, interleukin 13 (IL-13) or an effective part thereof, interleukin 15 (IL-15) or an effective part thereof, and granulocyte-macrophage colony stimulating factor (GM-CSF) or an effective part thereof; a hormone; and a heat-shock protein such as HSP70 or an effective part thereof, HSP90 or an effective part thereof, HSC70 or an effective part thereof, GRP94 or an effective part thereof, and calreticulin (CRT) or an effective part thereof.

14. The method according to ~~any one of claims 3-13~~ Claim 3, wherein the third moiety is of lipid nature, such as a palmitoyl group, a myristyl group, a farnesyl group, a geranyl-geranyl group, a GPI-anchor, and an N-acyl diglyceride group, or wherein the third moiety is a polyhydroxypolymer such as a polysaccharide.

15. The method according to claim 14, wherein the polysaccharide serves as a carrier backbone to which the amyloidogenic polypeptide and the foreign T cell epitope are separately bound.

16. The method according to claim 15, wherein the amyloidogenic polypeptide and the foreign T cell epitope are bound via an amide bond to the polysaccharide.

17. The method according to ~~any one of the preceding claims~~ Claim 1, wherein the amyloidogenic polypeptide or subsequence thereof has been modified so as to preserve B-cell epitopes which are not exposed to the extracellular phase when present in a cell-bound form of the precursor polypeptide for the amyloidogenic polypeptide.

18. The method according to claim 17, wherein the amyloidogenic polypeptide has been modified so as to lack at least one B-cell epitope which is exposed to the extracellular phase when present in a cell-bound form of the precursor polypeptide.

19. The method according to ~~any of the preceding claims~~ Claim 1 which comprises a substitution of at least one amino acid sequence within the amyloidogenic polypeptide with an amino acid sequence of equal or different length which gives rise to a foreign $T_H$ epitope in the analogue.

20. The method according to ~~any of the preceding claims~~ Claim 1, wherein the amyloidogenic polypeptide is selected from the group consisting of beta-amyloid (Aβ), amyloid precursor protein (APP), ApoE4, presenillin, a prion polypeptide, Alpha1-antichymotrypsin (ACT), Alpha2-macroglobulin, ABAD (Aβ-peptide binding alcohol dehydrogenase), APLP1 and -2 (amyloid precursor like protein 1 and -2), AMY117, Bax, Bcl-2, Bleomycin hydrolase, BRI/ABRI, Chromogranin A, Clusterin/apoJ, CRF (corticotropin releasing factor) binding protein, EDTF (endothelial-derived toxic factor), a heparan sulfate proteoglycans, human collapsin response mediator protein-2, huntingtin, ICAM-I, IL-6, lysosome-associated antigen CD68, P21 ras, PLC-delta 1 (phospholipase C isoenzyme delta 1), Serum amyloid P component (SAP), Synaptophysin, Synuclein (alpha-synuclein or NACE), TGF-b1 (transforming growth factor b1), full length or a fragment of V domain of IG light chain, a 76-residue N-terminal fragment of amyloid A protein, full-length or a fragment of transthyretin variants, an N-terminal fragment of ApoA1 variants, a full-length lysozyme variant, a 37-residue fragment of islet-amyloid polypeptide, a full-length wild-type insulin, full-length or a fragment of prion protein, a fragment of calcitonin, full-length or a fragment of transthyretin, full-length wild-type β-2 microglobulin, atrial natriuretic factor, a 110-residue fragment of variant cystatin, a 71-residue fragment of gelsolin variants, and a fragment of fibrinogen α-chain variants.

21. The method according to ~~any of the preceding claims~~ Claim 1, wherein the amyloidogenic polypeptide is (Aβ).

22. The method according to claim 21, wherein the amino acid sequence containing the foreign $T_H$ epitope is introduced into

the amyloidogenic polypeptide as schematically shown for the P2 and P30 epitopes in Fig. 1.

23. The method according to claim 22, wherein the amyloidogenic polypeptide comprises an amino acid sequence corresponding to amino acids 700-714 in SEQ ID NO: 2, such as an amino acid sequence consisting of amino acid residues 672-714 in SEQ ID NO: 2.

24. The method according to claim 23, wherein the amyloidogenic polypeptide comprises the amino acid sequence corresponding to amino acids 672-714 in SEQ ID NO: 2, wherein is inserted an amino acid sequence which gives rise to a foreign $T_H$ epitope in the analogue, or wherein the amyloidogenic polypeptide comprises an amino acid sequence corresponding to amino acids 672-714 of SEQ ID NO: 2, wherein at least one amino acid sequence is substituted by an amino acid sequence of equal or different length so as to give rise to a foreign $T_H$ epitope.

25. The method according to Claim 1 ~~any one of the preceding claims~~, wherein presentation to the immune system is effected by having at least two copies of the amyloidogenic polypeptide, the subsequence thereof or the modified amyloidogenic polypeptide covalently of non-covalently linked to a carrier molecule capable of effecting presentation of multiple copies of antigenic determinants.

26. The method according to Claim 1 ~~any the preceding claims~~, wherein the amyloidogenic polypeptide, the subsequence thereof, or the modified amyloidogenic polypeptide has been formulated with an adjuvant which facilitates breaking of autotolerance to autoantigens.

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27. The method according to any ~~one of the preceding claims~~ Claim 1, wherein an effective amount of the amyloidogenic polypeptide or the analogue of the amyloidogenic polypeptide is administered to the animal via a route selected from the parenteral route such as the intradermal, the subdermal, the intracutaneous, the subcutaneous, and the intramuscular routes; the peritoneal route; the oral route; the buccal route; the sublinqual route; the epidural route; the spinal route; the anal route; and the intracranial route.

28. The method according to claim 27, wherein the effective amount is between 0.5 µg and 2,000 µg of the amyloidogenic polypeptide, the subsequence thereof or the analogue thereof.

29. The method according to claim 27 ~~or 28~~, wherein the amyloidogenic polypeptide or analogue is contained in a virtual lymph node (VLN) device.

30. The method according to ~~any one of claims 1-21~~ Claim 1, wherein presentation of modified amyloidogenic polypeptide to the immune system is effected by introducing nucleic acid(s) encoding the modified amyloidogenic polypeptide into the animal's cells and thereby obtaining *in vivo* expression by the cells of the nucleic acid(s) introduced.

31. The method according to claim 30, wherein the nucleic acid(s) introduced is/are selected from naked DNA, DNA formulated with charged or uncharged lipids, DNA formulated in liposomes, DNA included in a viral vector, DNA formulated with a transfection-facilitating protein or polypeptide, DNA formulated with a targeting protein or polypeptide, DNA formulated with Calcium precipitating agents, DNA coupled to an inert carrier molecule, DNA encapsulated in chitin or chitosan, and DNA formulated with an adjuvant.

32. The method according to claim 31, wherein the nucleic acid(s) is/are contained in a VLN device.

33. The method according to ~~any one of claims 22-32~~ Claim 22, which includes at least one administration/introduction per year, such as at least 2, at least 3, at least 4, at least 6, and at least 12 administrations/introductions.

34. A method for treating and/or preventing and/or ameliorating Alzheimer's disease or other diseases and conditions characterized by amyloid deposits, the method comprising down-regulating amyloid according to the method of ~~any one of claims 1-33~~ Claim 1 to such an extent that the total amount of amyloid is decreased or that the rate of amyloid formation is reduced with clinical significance.

35. An analogue of an amyloidogenic polypeptide which is derived from an animal amyloidogenic polypeptide wherein is introduced a modification which has as a result that immunization of the animal with the analogue induces production of antibodies against the amyloidogenic polypeptide.

36. An analogue according to claim 35, wherein the modification is as defined in ~~any one of claims 1-19 and 22-24~~ Claim 1.

37. An immunogenic composition comprising

- an immunogenically effective amount of an amyloidogenic polypeptide autologous in an animal, said amyloidogenic polypeptide being formulated together with an immunologically acceptable adjuvant so as to break the animal's autotolerance towards the amyloidogenic polypeptide, the composition further comprising a pharmaceutically and immunologically acceptable carrier and/or vehicle, or

an immunogenically effective amount of an analogue according to claim 35 ~~or 36~~, the composition further comprising a pharmaceutically and immunologically acceptable carrier and/or vehicle and optionally an adjuvant.

38. A nucleic acid fragment which encodes an analogue according to claim 35 ~~or 36~~.

39. A vector carrying the nucleic acid fragment according to claim 38, such as a vector that is capable of autonomous replication.

40. The vector according to claim 39 which is selected from the group consisting of a plasmid, a phage, a cosmid, a mini-chromosome, and a virus.

41. The vector according to any claim 39 ~~or 40~~, comprising, in the 5'→3' direction and in operable linkage, a promoter for driving expression of the nucleic acid fragment according to claim 38, optionally a nucleic acid sequence encoding a leader peptide enabling secretion of or integration into the membrane of the polypeptide fragment, the nucleic acid fragment according to claim 38, and optionally a terminator.

42. The vector according to ~~any one of claims 39-41~~ Claim 39 which, when introduced into a host cell, is capable or incapable of being integrated in the host cell genome.

43. The vector according to claim 41 ~~or 42~~, wherein a promoter drives expression in a eukaryotic cell and/or in a prokaryotic cell.

44. A transformed cell carrying the vector of ~~any one of claims 39-43~~ Claim 39, such as a transformed cell which is capable of replicating the nucleic acid fragment according to claim 38.

45. The transformed cell according to claim 44, which is a microorganism selected from a bacterium, a yeast, a protozoan, or a cell derived from a multicellular organism selected from a fungus, an insect cell such as an $S_2$ or an SF cell, a plant cell, and a mammalian cell.

46. The transformed cell according to claim 44 ~~or 45~~, which expresses the nucleic acid fragment according to claim 38, such as a transformed cell, which secretes or carries on its surface, the analogue according to claim 35 ~~or 36~~.

47. The method according to ~~any one of claims 1-19 and 22-24~~, wherein presentation to the immune system is effected by administering a non-pathogenic microorganism or virus which is carrying a nucleic acid fragment which encodes and expresses the amyloidogenic polypeptide or analogue.

48. A composition for inducing production of antibodies against an amyloidogenic polypeptide, the composition comprising

- a nucleic acid fragment according to claim 38 or a vector according to ~~any one of claims 39-43~~, and
- a pharmaceutically and immunologically acceptable carrier and/or vehicle and/or adjuvant.

49. A stable cell line which carries the vector according to ~~any one of claims 39-43~~ and which expresses the nucleic acid fragment according to claim 38, and which optionally secretes or carries the analogue according to claim 35 ~~or 36~~ on its surface.

50. A method for the preparation of the cell according to ~~any one of claims 44-46~~, the method comprising transforming a host

cell with the nucleic acid fragment according to claim 38 or with the vector according to ~~any one of claims 39-43~~ claim 39.

51. A method for the identification of a modified amyloidogenic polypeptide which is capable of inducing antibodies against unmodified amyloidogenic polypeptide in an animal species where the unmodified amyloidogenic polypeptide is a self-protein, the method comprising

- preparing, by means of peptide synthesis or genetic engineering techniques, a set of mutually distinct modified amyloidogenic polypeptides wherein amino acids have been added to, inserted in, deleted from, or substituted into the amino acid sequence of an amyloidogenic polypeptide of the animal species thereby giving rise to amino acid sequences in the set which comprise T-cell epitopes which are foreign to the animal species, or preparing a set of nucleic acid fragments encoding the set of mutually distinct modified amyloidogenic polypeptides,

- testing members of the set of modified amyloidogenic polypeptides or nucleic acid fragments for their ability to induce production of antibodies by the animal species against the unmodified amyloidogenic polypeptide, and

- identifying and optionally isolating the member(s) of the set of modified amyloidogenic polypeptides which significantly induces antibody production against unmodified amyloidogenic polypeptide in the species or identifying and optionally isolating the polypeptide expression products encoded by members of the set of nucleic acid fragments which significantly induces antibody production

against unmodified amyloidogenic polypeptide in the animal species.

52. A method for the preparation of an immunogenic composition comprising at least one modified amyloidogenic polypeptide which is capable of inducing antibodies against unmodified amyloidogenic polypeptide in an animal species where the unmodified amyloidogenic polypeptide is a self-protein, the method comprising

- preparing, by means of peptide synthesis or genetic engineering techniques, a set of mutually distinct modified amyloidogenic polypeptides wherein amino acids have been added to, inserted in, deleted from, or substituted into the amino acid sequence of an amyloidogenic polypeptide of the animal species thereby giving rise to amino acid sequences in the set comprising T-cell epitopes which are foreign to the animal,

- testing members of the set for their ability to induce production of antibodies by the animal species against the unmodified amyloidogenic polypeptide, and

- admixing the member(s) of the set which significantly induces production of antibodies in the animal species which are reactive with the amyloidogenic polypeptide with a pharmaceutically and immunologically acceptable carrier and/or vehicle, optionally in combination with at least one pharmaceutically and immunologically acceptable adjuvant.

53. The method according to claim 51 ~~or 52~~, wherein preparation of the members of the set comprises preparing mutually distinct nucleic acid sequences, each sequence being a nucleic acid sequence according to claim 38, inserting the nucleic

acid sequences into appropriate expression vectors, transforming suitable host cells or host animals with the vectors, and effecting expression of the nucleic acid sequences, optionally followed by isolating the expression products.

54. The method according to claim 53, wherein the preparation of the nucleic acid sequences and/or the vectors is achieved by the aid of a molecular amplification technique such as PCR or by the aid of nucleic acid synthesis.

55. Use of an amyloidogenic polypeptide or a subsequence thereof for the preparation of an immunogenic composition comprising an adjuvant for down-regulating amyloid in an animal.

56. Use of an amyloidogenic polypeptide or a subsequence thereof for the preparation of an immunogenic composition comprising an adjuvant for the treatment, prophylaxis or amelioration of Alzheimer's disease or other conditions characterized by amyloid deposits.

57. Use of an analogue of an amyloidogenic polypeptide for the preparation of an immunogenic composition optionally comprising an adjuvant for down-regulating amyloid in an animal.

58. Use of an analogue of an amyloidogenic polypeptide for the preparation of an immunogenic composition optionally comprising an adjuvant for the treatment, prophylaxis or amelioration of Alzheimer's disease or other conditions characterized by amyloid deposits.

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